[1886.

#### HISTORY AND BIOLOGY OF PEAR BLIGHT.

#### BY J. C. ARTHUR.

To the American orchardist or nurseryman the name of pear blight, or fire blight, as it is often called, brings to mind a serious malady of fruit trees, which has been the theme of incessant discussion by horticultural writers and speakers since the earliest days of fruit culture in this country. The most marked features of the disease were admirably characterized by William Coxe<sup>1</sup> at the beginning of the present century, in the following words: "That species of blight which is sometimes called the fire blight, frequently destroys trees in the fullest apparent vigor and health, in a few hours, turning the leaves suddenly brown, as if they had passed through a hot flame, and causing a morbid matter to exude from the pores of the bark, of a black ferruginous appearance; this happens through the whole course of the warm season, more frequently in weather both hot and moist." The disease occurs from Canada and Minnesota on the north, to Georgia and Louisiana on the south, and from the eastern limit of the Rocky Mountains to the Atlantic ocean. No part of this vast extent of country is exempt, although it does not appear with the same frequency and power in all localities, and is usually rare in the immediate vicinity of the sea-coast.

So far as at present known, it is exclusively confined to this part of North America. This is partly inferred from the absence of any distinct mention of such a disease in the horticultural literature of other regions, and partly from direct testimony. Prof. Dwinelle, late of the University of California, has told the writer that it does not occur on the Pacific coast. Dr. De Bary,<sup>2</sup> whose word carries great weight, says, after giving a brief description of the disease, "this phenomenon is not to my knowledge known in Europe." A long account of the disease has been published by Dr. Wakker,<sup>3</sup> in a gardening journal of Holland, in order to learn if it occurs in that country, but up to the present

<sup>&</sup>lt;sup>1</sup> Cultivation of Fruit Trees, Philadelphia, 1817, p. 174.

<sup>&</sup>lt;sup>2</sup> Vorlesungen über Bacterien, 1885, p. 137.

<sup>&</sup>lt;sup>3</sup> Nederlandsche Tuinbouwblad, II (Jan. 9, 1886), p. 9.

time no one has intimated any knowledge of it. In a recent letter Dr. Masters, editor of the Gardeners' Chronicle of England, says that no such disease has been recognized in the British Isles. The testimony of one of our own horticulturists, Prof. Budd,1 of Iowa, who is familiar with the disease in this country, and has inspected the orchards of the old world far into Russia, is especially valuable; he says "no trace of blight of pear- or apple-trees can be seen in Europe." From these statements, and the inferences to be drawn from other sources, it appears highly probable that the disease does not extend to Europe. An account of the principal diseases of fruit-trees of New Zealand, by Prof. T. Kirk,<sup>2</sup> has been published, which describes a disease of the pear known in that country as fire blight, due to a fungus, and another of the apple, the American blight, due to an insect. No true pear blight, as recognized in the United States, is mentioned, and in a recent communication the author has definitely stated that it is not known in the colony. Whether it occurs in other parts of the world is not yet ascertained, if some slight testimony regarding its absence in Japan be excepted.

It is only within a year or so that European writers have become aware of its existence, and this only through American authors. It is remarkable that a disease of such virulence and so easily transported should not have found its way across the ocean, when one remembers the number of destructive plant maladies that America has already involuntarily foisted upon European cultivators. It will not be profitable to speculate much at this time upon the reasons for this, but we may suppose that the small exportation of American fruit-trees, or of scions,<sup>3</sup> has been a factor in keeping it in check. The influence of climate, and some less evident factors, need not be discussed in this connection.

Amount of Loss.—It has already been intimated that pear blight is a frequent and destructive affection; it will tend to give a fairer appreciation of the subject if it be stated how frequent and how destructive it is. Coxe,<sup>4</sup> as early as 1817, in the oldest pomological work by an American author, says it "frequently

<sup>&</sup>lt;sup>1</sup> Trans. Minn. Hort. Soc. for 1883, p. 281.

<sup>&</sup>lt;sup>2</sup> Fruit Blights and Diseases of Fruit-trees in New Zealand, 1885.

<sup>&</sup>lt;sup>3</sup> For an account of the destruction of stored scions by blight, see Rep. Hort. Soc. of Mich. for 1881, p. 137. <sup>4</sup> L. c., p. 174.

destroys trees in the fullest apparent vigor and health, in a few hours; I have in twenty years lost upwards of fifty trees." The years 1826 and 1832 were notable in horticultural circles for the increased prevalence of the disease; but it was in 1844 that the most widespread and fatal epidemic, that the country has yet known, occurred. Few, if any, pear-orchards escaped at that time without the partial or total loss of many trees, and some orchards, even large ones, were quite destroyed. The following year the epidemic was much lighter, and had fully disappeared by 1846. Although it had subsided as an epidemic, it still occurred in localities here and there, and has continued to do so until the present time. Judging from the communications in the horticultural press, the whole country, or various sections of it independently, have been subject at various times to epidemic visitations, but none have equaled in severity that of the memorable year of 1844.

It is often maintained that a certain periodicity of occurrence is observable, the periods usually being placed at five, ten, or twenty years. A careful examination of the literature of the subject, however, gives little support to these views, and makes it more probable that the intervals are irregular, and that they vary for different sections of the country. The year of maximum prevalence may or may not be preceded by one in which the disease is noticeably common, but it is quite invariably followed by a year or two of successive decadence.

In the absence of exact statistics, which it has not been practicable to obtain, something of the important nature of the disease may be gathered from the statements of horticultural writers and the phraseology which they employ in speaking of it.

The renowned horticulturist, A. J. Downing,<sup>1</sup> called it the "monstrous malady of the pear." Chas. R. Baker<sup>2</sup> says it is "the worst malady with which the cultivator of the pear-tree has to contend." In southern Pennsylvania "the pear is so generally destroyed by the blight," according to J. B. Garber<sup>3</sup> writing in 1850, "that very few trees are to be found." At Philadelphia, however, the disease has been rarely observed, according to

[1886.

<sup>&</sup>lt;sup>1</sup> Horticulturist, vol. i, 1846, p. 62.

<sup>&</sup>lt;sup>2</sup> Practical and scientific Fruit Culture. Boston, 1866, p. 476.

<sup>&</sup>lt;sup>3</sup> U. S. Patent Office Report for 1850, pt. ii, p. 418.

Thomas Meehan.<sup>1</sup> T. T. Lyons,<sup>2</sup> of Michigan, states as the opinion of many cultivators in that State, that the pear-tree cannot be grown with financial success on account of the blight. Illinois has always been much subject to the disease, and Prof. J. B. Turner,<sup>3</sup> in 1868, gave expression to the general feeling of his region by describing it as "that deadly Upas of the pear-tree known par excellence as the pear-blight." In 1882 Dr. J. L. Hallum,<sup>4</sup> speaking for southern Illinois, says, " pears have failed, utterly failed, so that none are now cultivated for market, the blight has destroyed the trees-branch and root," and S. G. Minkler,<sup>5</sup> in the northern part of the State, observes that it is a very uncommon thing to see pear-trees without dead branches or other signs of the ravages of blight. Wm. A. Nourse,6 of the same State, is led to "doubt if one-tenth of the pear-trees that are set, live ten years," on account of this one destructive agent. Geo. M. Dewey,<sup>7</sup> of Missouri, says that "with good cultivation and rich soil the pear generally dies of blight before the eighth year." In Minnesota the severe climate has not permitted the cultivation of pears, and almost the only apples grown for many years were the hardy crab-apples. The latter have been rapidly improved, and together with the hardier varieties of the common apple would now furnish this part of the country with an abundant supply of fruit, were it not for this same disease, which elsewhere most conspicuously preys upon the pear-tree. E. H. S. Dartt<sup>8</sup> held the opinion in 1874 that the severity of winter was not so much to be dreaded as the ravages of blight. He had at that time one or two thousand trees affected. Dr. P. A. Jewell,<sup>9</sup> up to 1876, had lost ten thousand Tetofsky apple-trees by it. F. G. Gould 10 says that " only for this scourge every family living on a farm in Minnesota could have a supply of apples."

- <sup>4</sup> Same for 1882, p. 118.
- <sup>5</sup> Same for 1880, p. 30.
- <sup>6</sup> Trans. Ill. Hort. Soc. for 1880, p. 63.
- <sup>7</sup> Proc. Mo. Hort. Soc. for 1870, p. 18.
- <sup>8</sup> Trans. Minn. Hort. Soc. for 1874, p. 22.
- <sup>9</sup> Same for 1876, p. 73.
- <sup>10</sup> Same for 1884, p. 127.

<sup>&</sup>lt;sup>1</sup> Rep. Penn. Fruit-Grower's Soc. for 1877, p. 77.

<sup>&</sup>lt;sup>2</sup> Rep. Pomol. Soc. of Mich., for 1878, p. 368.

<sup>&</sup>lt;sup>3</sup> Trans. Ill. Hort. Society for 1868, p. 42.

**[1886.** 

Citations enough have doubtless been given, although several pages of equally strong ones might be added, to show that fruitgrowers, who have the best opportunities for observation, consider it a disease greatly to be dreaded and one of special economic importance. Other sections of the country, notably those of Ohio, western New York and Georgia, could furnish equally important proof of these propositions. All that is desired in this connection, however, is to give those not familiar with the subject some idea of the disease and its effects as ordinarily observed

Early Records.—The oldest mention of the disease, that gives a good and reasonably full description of it, is in Coxe's work on fruit-trees, bearing the date of 1817. The manner of the author leaves no doubt that it was well known at that time, and the reference to his losses during twenty years makes it reasonably certain that he had observed the disease as early as the opening of the century. The earliest notice, however, which has yet come to hand, is in a letter written by Wm. Denning,<sup>1</sup> describing the disease in apples, pears, and quinces. He speaks of first observing it on the Highlands of the Hudson in 1780.

There is no interest, however, in tracing chronologically the various notices found in different publications, for without exception they have the tone of treating a familiar theme, and show no evidence that the disease in the first part of the century was in any respect different from to-day.

Conjectures Regarding its Cause.—A brief treatment of this topic will be all that is required for the purposes of this paper; and only those hypotheses will be touched upon which received such careful presentation as to attract the favorable attention of the public.

Few writers appear to ascribe the disease to a single agency, but regard it as resulting from several causes, either acting together or brought about by dissimilar circumstances. Little discrimination is made between predisposing conditions and active agents. In fact sharply defined treatment could not be expected when all was conjecture, and when the shrewdest observers did not hesitate to avow that after years of loss under all kinds of experimentation, and after interminable discussions, the cause still lay shrouded in impenetrable obscurity.

<sup>&</sup>lt;sup>1</sup> Trans. Soc. for Prom. of Agric., vol. ii, 1794, p. 219.

Coxe,<sup>1</sup> who has had many followers, thought that the hot rays of the sun when acting through a misty or saturated atmosphere deranged the vital activities of the plant and brought about the disease. He considered old varieties more subject to it, on account of having lower constitutional vigor, than new varieties, of which the St. Germain and Seckel were respectively conspicuous instances.

The insect theory, as it was called, was promulgated at this time. It was started upon firm facts by the discovery of a small brown beetle, about two millimeters long, which penetrated the branch, and caused the part beyond to die. The beetle received the name of Scolytus pyri Peck, now changed to Xyleborus pyri Pk., and is still known as the blight beetle. The effect of its attack appears to the casual observer similar to that of the true blight-the branch in June or July rapidly withers, and the leaves and fruit turn black. The beetles being minute and inconspicuous escape attention, and the fact that the branch does not die below a definite point is sometimes overlooked. It is not difficult to see how many persons came to connect this comparatively rare affection with the common fire blight, and to believe that insects of some sort were to be held accountable for all-their supposed minuteness and wary habits being sufficient reasons for the failure to find them, and the spread of the disease along the limbs of a tree being ascribed to a poison which the insects were supposed to emit. Among the prominent supporters of this view was the "Genesee Farmer,"<sup>2</sup> published at Rochester, N. Y., with Patrick Barry as the horticultural editor. It has not, however, been so strongly advocated for the last decade or two.

The next hypothesis that attracted general attention was known as the frozen-sap theory. This was based upon the supposition that the autumn or winter freezing of unripe wood produced a poison which the moving currents of sap the next spring and summer distributed, causing the death of the parts. It was first published in 1844 by Rev. H. W. Beecher,<sup>3</sup> of Indiana, in a long and able article in "Hovey's Magazine." In the following year

327

<sup>&</sup>lt;sup>1</sup> L. c., p. 175.

<sup>&</sup>lt;sup>2</sup> See Genesee Farmer, vol. vii, 1846, p. 216; vol. viii, 1847, pp. 122, 218, etc.

<sup>&</sup>lt;sup>3</sup> Magazine of Horticulture, vol. x, p. 441.

it was independently elaborated by A. J. Downing<sup>1</sup> in his work on Fruits and Fruit-trees of America, who first called it the frozen-sap theory, and who is usually spoken of as the author of it. This view has probably had more firm adherents than any other, as it explained many phenomena connected with the disease in a fairly satisfactory manner. It was especially well received in the western States.

The next hypothesis which gained the attention of the public was the fungus theory. Its first successful presentation was in 1863 by Dr. J. H. Salisbury,<sup>2</sup> who figured the fungus which he decided to be the specific cause of this kind of blight, and ventured to give it a name, although he was sadly in error in most that he did. Thomas Meehan,<sup>3</sup> editor of the "Gardener's Monthly," has ably championed this explanation, and done much to keep it in favor. In 1875, Dr. J. G. Hunt,<sup>4</sup> by Mr. Meehan's request, undertook a microscopical examination of blighted pear-twigs, and confirmed the opinion that it was due to a fungus, without, however, deciding upon the specific character of it.

Blighted trees often attract attention immediately after a thunder storm, and from this and other circumstances the belief that the malady is due to electricity has gained many adherents, but the argument has not had a full and connected presentation.

The last hypothesis of historical importance is the bacterial theory. Although hinted at by a number of horticultural writers, yet the credit of it is due to Prof. T. J. Burrill,<sup>5</sup> who in 1878 distinctly stated his belief that the cause resides with the bacteria which he found in great abundance in the tissues of affected branches. In 1880 he performed a series of experiments <sup>6</sup> by inoculating healthy branches with the juices of diseased ones, the results of which were presented to the American Association at its Boston meeting, thus first bringing the subject clearly to the

<sup>1</sup> Fruits and Fruit-trees of America, p. 594; same, 2d Revision by Chas. Downing, p. 646.

<sup>6</sup> Proc. Amer. Assoc. Adv. Sci., vol. xxix, 1880, p. 583; Rep. of Ill. Industrial Univ., for 1880, p. 62; Trans. Ill. Hort. Soc. for 1880, p. 157; Amer. Naturalist, vol. xv, 1881, p. 527.

<sup>&</sup>lt;sup>2</sup> Ohio Agric. Rep. for 1863, p. 450.

<sup>&</sup>lt;sup>3</sup> Proc. Amer. Pomol. Soc. for 1867, p. 59; and elsewhere.

<sup>&#</sup>x27; Gardener's Monthly, vol. xvii, 1875, p. 245.

<sup>&</sup>lt;sup>5</sup> Trans. Ill. Hort. Soc. for 1878, p. 80.

attention of the scientific world. Although this was now the popular hypothesis, it cannot be said to have received more substantial credence than those which had gone before, either from the horticulturists or the scientists. The experimental results gained by Prof. Burrill were confirmed and extended by the writer <sup>1</sup> during 1884, by means of a similar series of inoculations.

Of the multitude of minor hypotheses which were put forth in explanation of phenomena connected with pear-blight, and which were variously received, and of all degrees of plausibility, it is impossible to speak at this time without carrying this paper to undue length.

Beginning of Experimental Research.—The question of the cause of pear-blight was finally removed from the domain of speculation to that of established fact by a series of crucial experiments performed by the writer<sup>2</sup> a year ago, and recorded in a paper before the American Association at the Ann Arbor meeting. These consisted in showing that the bacteria when removed from the tree and passed through a series of artificial cultures would generate the disease when again introduced into the tree, and that the juices accompanying blight when cleared of bacteria by filtration will not produce the disease.

Having now come to a firm basis for scientific advancement, let us look over the historical ground again to see if some one did not hit upon the true explanation of the disease, although he may not have been aware of its significance. In a connection like this, facts derived from experiment have greater weight than statements of opinion; the latter acquire importance in proportion as they are logically derived from correct and close observation. Bearing this in mind, we need not give much heed to the not uncommon inference that pear-blight was in some way intimately related to the epidemic diseases of man, e. g. cholera. This view is said to have been quite frequently entertained in the early part of the century, but was not sanctioned by the learned. The use of such phrases as "first cousin to the cholera," "a species of vegetable ferment," etc., surely does not entitle the author to any priority in way of discovery.

<sup>&</sup>lt;sup>1</sup> Rep. N. Y. Agric. Exper. Station for 1884, p. 357.

<sup>&</sup>lt;sup>2</sup> Proc. Am. Assoc. Adv. Sci., vol. xxxiv, 1885, p. 295; Bot. Gazette, vol. x, 1885, p. 343; Gardeners' Chronicle, vol. xxiv. 1885, p. 586.

We turn from these slight hints to the record of an experiment in inoculation, made in 1845 and published the following year. We are told by S. B. Gookins,<sup>1</sup> of Indiana, that visiting Mr. Ragan (the same person who furnished Mr. Beecher with many of the facts on which he founded the theory to which we have already referred) he was shown a thrifty young pear-tree in the nursery, which had been "inoculated" "by way of experiment" with "the sap of a blighted tree," "a few days previous." "He made an incision about three feet from the ground, lifted the bark as in the process of budding, and injected a small quantity of the diseased sap." "We found the leaves of the patient changing color, and emitting that peculiar odor which is always present in cases of blight, and upon applying the knife, the inner bark was found to be black from the root to the top, while nothing of the kind appeared elsewhere in the nursery."

This admirable experiment was combined with a no less admirable interpretation of the cause of blight. The writer cites facts to disprove the hypothesis of Mr. Beecher, and then says: "I strongly incline to the belief, that the pear-blight is an epidemic, that it prevails like other epidemics, and will pass off like them. The atmosphere is, I believe, generally admitted to be the medium by which they prevail, and are carried from place to place. What that subtle principle may be, which pervades our atmosphere, by which infection is retained and transmitted, human science has not discovered; but that such a principle exists is sufficiently obvious from its effects."

This clearly conceived elucidation of the matter could only have been improved by a knowledge of the germ theory of disease, and when we remember the date at which it was uttered, we do not feel that the writer was guilty of any lack of acuteness in not perceiving the relation which we now know to exist between his theory and his facts. He seems to have been a modest man, for he only signs his initials, and does not defend his views when the editor, A. J. Downing,<sup>2</sup> opposes the opinion that "an epidemic conveyed by the atmosphere is too slightly supported by facts to weigh at all against the observations of cultivators," which "strongly point to the freezing of the sap as the cause."

<sup>&</sup>lt;sup>1</sup> Horticulturist, vol. i, 1846, p. 253.

<sup>&</sup>lt;sup>2</sup> Horticulturist, vol. i, 1846, p. 255.

Another interesting experiment was performed by Dr. E. S. Hull,<sup>1</sup> of Illinois, in 1868. Having received some blighted apple twigs from a correspondent, he cut pieces from them with which he "inoculated several succulent pear shoots by tying in the pieces as in budding." This was done the middle of June, and no observation taken for thirty-four days, when the blight was found to have extended several inches into the healthy tissues. From this he very justly concludes that the blight in apples and pears is but one disease, but seems to take it for granted that in both it is due to "vitiated sap."

The fact that the disease may be transferred to healthy trees by the pruning knife has been observed by several persons. H. Wendell,<sup>2</sup> of New York, says in 1849, "I am also careful that the blade of the knife is perfectly clean, and that it has none of the sap of a diseased tree adhering to it, because I have known many valuable trees destroyed by having been inoculated in this manner." Prof. Turner,<sup>3</sup> of Illinois, makes a similar statement: "I found that this disease is exceedingly contagious, for if I used my knife to prune a healthy tree after having used it in shaving the diseased one, I communicated the disease to that tree."

Prof. Burrill first observed the bacteria of blight in 1877,<sup>4</sup> but did not recognize them as such till the following year,<sup>5</sup> when he avowed his belief that they were the cause of the disease. His first inoculation experiments were made in 1880, as already stated. In 1882 he characterized the organism under the name of *Micrococcus amylovorus*.<sup>6</sup>

Description of Micrococcus amylovorus Bur.— The form of this species of bacteria is very constant, under all conditions. The single cells are from oval to roundish-ovoid, and only vary by slight changes in the ratio between their length and breadth

<sup>5</sup> Same for 1878, page 79.

<sup>6</sup> The Bacteria (a reprint from Rep. of Ill. Industrial Univ. for 1882), p. 42; Amer. Naturalist, vol. vii, 1883, p. 319. In the last publication, by a typographical error, the name was made to read *M. amylivorus*, a mistake which has been copied into other works—see Grove's Bacteria and Yeast Fungi, London, 1884, p. 10.

331

<sup>&</sup>lt;sup>1</sup> Trans. Ill. Hort. Soc. for 1868, p. 220.

<sup>&</sup>lt;sup>2</sup>U. S. Patent Office Rep. for 1849, pt. ii, p. 446.

<sup>&</sup>lt;sup>3</sup> Trans. Ill. Hort. Soc. for 1878, p. 81.

<sup>\*</sup> Same for 1877, page 114.

(Pl. III, figs. 1, 2, 6). They are 1 to  $1\frac{1}{4}\mu$  long, by  $\frac{1}{2}$  to  $\frac{3}{4}\mu$  broad, and quite colorless. For the most part, they exist as single independent cells, but may often be found in pairs, especially when still multiplying, and in rare instances are united into a series of four or even more, but never extend into chains.

During rapid vegetation, in rich nutritive media, the movements reach a stage of extreme activity. The appearance is what is termed swarming, in which the bacteria move rapidly back and forth, in and out among each other, but never in a straight line to any distance. As the rate of growth becomes less from any cause the movements are retarded. Taken directly from the tissues of a blighting tree, the movements of translation are usually sluggish or imperceptible, although the universal Brownian movement is likely to give a misleading appearance of activity. Under specially favorable conditions, as when grown during hot weather in very succulent shoots, or from artificial inoculation in unripe fruit, the movements are much increased and may become quite rapid. When taken from the tree in winter, or when grown in solutions that are too acid or too alkaline, or which are deficient in the proper nutritive substances, there is no perceptible locomotion.

When in active growth, the cells present a uniformly dull appearance. By conditions which are unfavorable to normal growth, yet do not entirely check it, such as strongly acid or alkaline solutions, deficient nutriment, or exhaustion by keeping the cultures several months, the cells become highly refractive, and to some extent take on the appearance of the spores of other species of bacteria. Whether in this state they possess any of the characteristic powers of resistance which belong to spores, has not been ascertained.

Formation of Zooglæa.—By far the most characteristic feature in the life history of *Micrococcus amylovorus* is the formation of zooglæa (figs. 2, 3, 5). These have never been observed in the tissues of the tree under any conditions, or in or upon any sort of solid media, but they occur with much regularity in fluid cultures, when placed under favorable conditions for rapid growth.

They are produced to some extent throughout the fluid, but are most abundant in the thin pellicle which forms upon the surface, appearing within forty-eight hours from the beginning

**[1886.** 

1886.]

of the culture. The substance of the pellicle consists of a colorless matrix uniformly filled with motionless bacteria, and against this the zooglea are sharply defined. They are often brought out yet more distinctly by being surrounded by a colorless layer, free of bacteria, which is doubtless an extension of the ground substance of the zooglea mass (fig. 4).

The masses are far more dense than the pellicle, and are compactly filled with refractive bacteria. They possess a definite outline, and are recognizable when very small; and although they may reach 30 to 40  $\mu$  long by 20 to 30  $\mu$  wide, they rarely lose their distinctness. When below 10  $\mu$  in length, their usual form is oblong, varying to globular. They occur singly, or united more or less intimately end to end in pairs, and sometimes several form a short chain. At this stage they possess a uniformly even and unbroken surface, which now becomes uneven and wrinkled, and is finally thrown into folds, giving some resemblance to the external aspect of the brain. Zooglea more than 20  $\mu$  in length have the folds of somewhat unequal height, and the sinuses deeper, giving a stronger cerebric look, or when the folds are small and circular, they are better described as mulberry-like. The elongated forms, which at some stage of growth might doubtless have been composed of two or more distinct masses, often take on a vermiform appearance. But whatever the variations may be, the distinctness of outline, the general form, and the cerebric surface are unfailing characters, which so far as my knowledge extends, are not found in any other species of bacteria.

Cultivation in Fluid Media.—The range of substances which may serve as culture media for this organism is very wide. An infusion of almost any vegetable substance containing a fair amount of soluble carbohydrates is likely to be sufficient to enable growth to take place, even if not very luxuriantly.

The substance which on the whole has proved most satisfactory is an infusion of potato. This is prepared by paring a potato and slicing it into three or four times its bulk of water. This is kept for a couple of hours at about  $70^{\circ}$  C., by placing it over a water-bath, during which time it is occasionally stirred. It is then filtered, and is ready to be placed in the culture vessels for sterilizing and use. If the heat is allowed to rise much above  $70^{\circ}$ , the starch is gelatinized, and it is only with difficulty that the solution can be filtered. The resulting liquid is clear and watery, but is often

333

light brown from coloring matter contained in the potato, which does not, however, materially interfere with observations on the growth of bacteria in it. Iodine gives a blue coloration to this liquid, showing that it contains starch, probably in the form of amylon. Another equally good culture fluid is made by treating corn (maize) meal in a similar manner. The solution is colorless, but it is very apt to throw down a troublesome sediment, which makes it less desirable to use than the potato solution.

Test-tube or flask cultures with these liquids, when kept at a temperature of 25° to 30°C., usually show some turbidity in twenty-four hours after being infected, and if the growth is very rapid, bubbles of gas  $(CO_2)$  will be given off, which collect at the surface into a slight froth. In forty-eight hours the liquid has become thoroughly turbid. By this time a thin whitish pellicle has formed on the surface, which does not increase much in thickness up to the end of active growth, and rarely becomes wrinkled. With the formation of the pellicle, a sediment gathers at the bottom of the liquid, often a centimeter in depth, but which is so light that it only apparently differs from the liquid above by being whiter. In the course of some weeks this sediment will mostly gather upon the bottom of the vessel. No difference has been observed in the appearance of the bacteria taken from different parts of the culture. Those imbedded in the pellicle are not arranged in any recognizable order.

In proportion as liquids are less suitable to the growth of the organism, the visible changes are less. The pellicle may not be formed, and there may be no turbidity, but if any growth at all takes place there will be some evidence of it by formation of a slight sediment. But the occurrence of a precipitate does not necessarily imply growth, for it not infrequently separates from a liquid containing organic matter, although remaining perfectly sterile.

An infusion of hay, and also of dead, partly decomposed grass from a marsh, gave nearly a normal growth of blight bacteria, but the cells were considerably more refractive than usual.

A solution of starch, having one part of starch to fifty of water, gave but a slight growth of highly refractive bacteria, without a pellicle, turbidity, or zooglæa. A strong decoetion of old barnyard manure acted in the same manner. A solution of one part of glucose to lifty of water gave no growth of the bacteria.

In testing the effect of acids upon the development of blight bacteria, a  $\frac{1}{2}$  per cent. of malic acid was added to the usual infusion of potato. This prevented the formation of a pellicle, turbidity or zooglea, but gave a very considerable cloudy sediment, largely made up of loosely aggregated groups of blight bacteria, which were brilliantly refractive. A similar solution with 2 per cent. of malic acid gave a slightly less abundant sediment, but with otherwise the same results. Some of the latter was transferred to a corn-meal solution, producing the characteristics of a pearblight culture, except the formation of zooglea. After some days this was introduced into a pear tree, which in due course of time gave the true blight, showing that the bacteria of the acid solution were really blight bacteria. Attempts to grow them in a nutrient 5 per cent. solution of citric and tartaric acids have not been successful.

Testing the nature of the bacteria in cultures producing limited growth, by inoculating directly into the tree, has not, as a rule, proved successful, as for some reason they seem unable to gain a footing in the living tissues. It is therefore necessary to transfer them first to richly nutrient cultures, from which, after a time, they may be introduced into the tree, and, if the blight bacteria are present, will start the disease.

Cultivation in Solid Media.—In test-tube cultures with nutrient gelatine the most characteristic results have been obtained by adding a drop containing blight bacteria to the gelatine while liquid, and thoroughly distributing the germs by shaking the tube. In from two to three days the gelatine contains numerous small white dots, which, upon examination under the microscope, prove to be a mass of bacteria of the usual appearance. The dots are globular or oval, and increase to about .5 mm. in diameter. No further growth or change takes place, and in this condition they remain for weeks, without liquefying or otherwise affecting the gelatine.

When sown upon the surface of the gelatine by drawing a needle or glass rod over it, or by placing a drop on it, the growth is feeble and does not amount to more than a slight shining appearance of the surface.

A nutrient solution made from an unripe pear, in which blight

germs were well distributed by shaking and then left undisturbed for two days, gave the same isolated white dots as in gelatine; but they dropped to the bottom of the liquid upon being jarred. The bacteria were evidently prevented from moving freely by the jelly, which was not, however, thick enough to keep the groups in place when its cohesion was once disturbed. Fruit jellies, doubtless, may be found to be convenient media for the cultivation of this species of bacteria.

No success has been attained in the use of agar agar, but whether due to a want of adaptability in the substance, or to wrong manipulation, must be left to future experiments to determine.

The opaque solid cultures proving most successful have been conducted upon freshly gathered unripe pears. Slices of these are placed under a moist bell-jar, and infected by touching with a needle that has been dipped in some substance containing the bacteria. In two or three days fine milky drops, like beads of dew, will appear scattered over the surface for 5 mm. or more about the infected spot. These will become somewhat larger after a time, while the spot which received the infection will turn slightly brown, the tissues gradually wasting away and forming a small depression.

If, however, the slices, having freshly cut surfaces both above and below, are laid upon a plate with a little water, and placed under a bell-jar, the result is not the same. The dew-like drops appear within forty-eight hours, as in the other case, but increase rapidly in size, while a drop is also formed at the point of infection. Drops finally appear over the whole surface of the slice. They remain more or less distinct, and soon become as large as a pea, retaining the globular or rounded form to a remarkable degree. Microscopically they are composed of the usual form of blight bacteria, suspended in a colorless fluid. After about a week, the drops coalesce and the tissues of the pear begin to break down. This sort of culture requires no precautions of sterilizing, as no other bacteria can multiply upon it till after the cells of the pear begin to die.

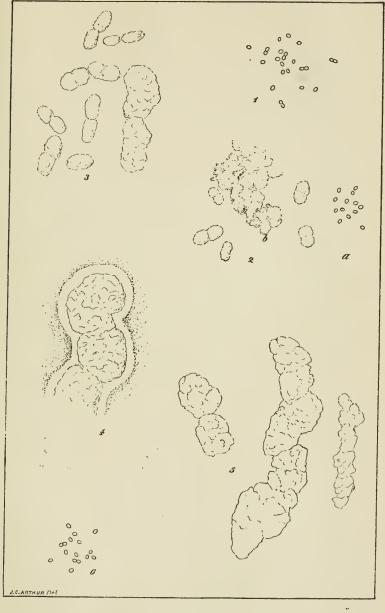
When blight bacteria are sown upon slices of baked or boiled potato, they spread out over the surface in a thin, slightly moist layer, which is usually somewhat yellowish, but do not grow

336

[1886.

# PROC. ACAD. NAT. SCI. PHILA., 1886.

# PLATE III.



MICROCOCCUS AMYLOVORUS BUR.



readily, or produce a characteristic appearance. Under the microscope the cells are strongly refractive.

337

A boiled potato was infected by thrusting a platinum wire, smeared with blight bacteria, into one end. After sixteen days it was cut open. No external change had taken place, and, to the unaided eye, no internal change either; the odor and texture were still those of a freshly boiled potato. The microscope, however, revealed the blight bacteria in every part of the potato, in irregular motionless masses, and with more than the usual refractiveness.

These opaque solid cultures have brought out one fact very distinctly, which is, that *Micrococcus amylovorus* requires a large supply of water for its best development—a fact which has an economic bearing.

Behavior toward Staining Fluids.—So far as trial has been made, nothing especially characteristic has been detected to distinguish this form of bacteria from the majority of micrococci. The most successful results have been obtained with a watery solution of Bismarck-brown, especially in cover-glass preparations. These make excellent specimens when mounted in Canada balsam.

The zooglæa are inclined to be too deeply stained by this process, and for most purposes they are best studied unstained. They may be well preserved by mounting in glycerine.

Hæmatoxylin has also given good results, but has not been found particularly useful.

Chemical Products.—The chemical changes brought about by the activity of the blight bacteria have not yet been fully and carefully worked out. The most obvious product is carbon dioxide, which often passes off so freely from a cultivation as to produce a slow effervescence. Butyric acid and alcohol are formed in very small quantities, if at all. The tests by which these facts have been determined have already been published,<sup>1</sup> and need not be repeated here. Vigorous cultures of the bacteria in infusion of potato give no reaction for glucose with Fehling's solution; and blighting tissues from the tree give no indication by the same test of more than the normal amount of glucose to be found in healthy tissues. On the other hand a quantitative

<sup>&</sup>lt;sup>1</sup> Rep. N. Y. Agric. Exper. Station for 1885, p. 247; and less fully in Amer. Nat., vol. xix, 1885, p. 1181.

determination of sound and blighting pears, taken from the tree at the same time, shows considerably less sugar in the latter.

A favorite explanation with horticulturists of the action of fireblight upon the pear-tree, has been to say that the sap is poisoned. This poison was supposed to be introduced by insects, or to be due to some disorganization of the tissues. Although it is now known that specific bacteria are directly answerable for the disease, it is yet worth while to see if the old idea of a poison has not some foundation in fact.

It has been ascertained that certain bacteria produce, during their growth, characteristic poisons which are classed under the name of ptomaines. Most of the ptomaines are non-volatile, and readily soluble in water or alcohol. The chemical tests which are applied for their detection cannot be considered conclusive except when taken collectively. The tests tried below are among the most satisfactory known at present.<sup>1</sup>

A cultivation in infusion of potato, giving about 200 cc. of liquid was filtered, and the filtrate evaporated to a syrup. This was treated with alcohol, and the solution tested with the most characteristic test for ptomaines—the reduction of potassic ferriccyanide. Other portions of the solution were successively tested with phospho-molybdic acid, potassio-mercuric iodide, and iodine in potassic iodide, all of which failed to give any distinctive reactions.

Another trial was made with about 200 cc. of material prepared by cooking a potato in just enough water to cover it, sterilizing, and cultivating the bacteria in it as usual. In four days from beginning of the culture it was filtered; the residue upon the filter was treated with 100 cc. of distilled water, slightly acidulated with hydrochloric acid, heated to  $70^{\circ}$ C. and filtered. The two filtrates were united and evaporated to a syrup. This was digested in the cold with alcohol containing a little sodic hydrate. This solution was tested as before, and also with platinic chloride and concentrated sulphuric acid, and all with no distinctive reactions.

A third trial was made with a boiled potato, which had been permeated with the blight. The extract was made by the Stas-Otto method, and the same reagents used as in the last case, with equally negative results.

<sup>&</sup>lt;sup>1</sup> Cf. Brieger, Ueber Ptomaine, 1885, p. 22, et seq.

These tests do not cover the possibility of the ptomaine being volatile, which is really not very great. It is yet necessary to make tests of freshly blighted tissues from the tree, which can only be done during the hot months.

Action of the Organism in the Living Plant.—The bacteria of blight have the power of growth and multiplication in the presence of the living cells of the pear, and in this one important respect differ essentially from other species of bacteria. By artificial inoculation into growing unripe pears, which give most marked and certain results, it is found that other bacteria are entirely innocuous, at once disappearing without having made any growth or induced any changes in the tissue of the pear. If blight bacteria in active condition are intermixed with the other forms, they penetrate the cells, multiply, and finally bring about the disorganization and death of the tissues which marks the progress of the disease, but the associated forms disappear the same as when introduced alone, and the product is a mass of practically pure blight bacteria.

This result is rendered possible on account of the fact already stated, that the blight bacteria penetrate the tissues, and maintain their normal growth for some time (days or weeks), before the life of the cells is sufficiently interfered with to permit the growth of other forms. The bacteria always extend beyond the visible location of the disease—in small branches, often to the distance of a third of a meter or more.

One of the properties which enables this species to successfully penetrate the pear-tree is evidently its unusual indifference to acids, which prevents most other forms from making any growth; the juices of the pear give a strong acid reaction with test paper.

What chemical changes are brought about by its activity in the plant cannot be definitely stated, further than to say that a mucilage or gum, which is soluble in water, is produced in abundance, with the disengagement of carbon dioxide. The contents of the cells, together with the cell-walls which have not been liquefied or changed into stony tissue, pass over into this viscous product.<sup>1</sup>

<sup>&</sup>lt;sup>1</sup> Rep. N. Y. Agric. Exper. Station for 1885, p. 248; Amer. Nat., vol. xix, 1885, p. 1181.